

## INFECTION OF *CERATIUM FURCA* BY THE PARASITIC DINOFLAGELLATE *AMOEBOPHRYA CERATII* (AMOEBOPHRYIDAE) IN THE MEXICAN PACIFIC

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### ABSTRACT

Parasitism within dinoflagellates is a widespread and well-documented phenomenon. Parasitic dinoflagellates of the genus *Amoebophrya* commonly infect free-living toxic, and non-toxic dinoflagellates species which may cause harmful red tides. Infections of *Ceratium furca* by *A. ceratii* were observed in red tides samples collected in the northwest coast of Baja California between 30°01'05" N, 115°51'16" W and 31°09'33" N, 116°31'09" W. This is the first record of this particular parasitic dinoflagellate in Mexican Pacific waters. There were mainly three dinoflagellate species causing this particular seawater discoloration: a *Gymnodinium*-like dinoflagellate, *Ceratium furca*, and *Akashiwo sanguinea*. These reached concentrations as high as 560 000, 762 600, and 395 400 cells L<sup>-1</sup>, respectively. During the bloom, surface water temperature ranged between 13 and 17°C. Seawater salinity ranged from 33.2 to 33.8 psu. About 1.5% of the individuals of *C. furca* observed were infected by the intracellular parasite dinoflagellate *Amoebophrya ceratii*. This parasite was observed mainly inside specimens of *Ceratium furca* and very few specimens of *Ceratium macroceros*. In general, individuals of *C. furca* were partially or totally deformed. Infections by *A. ceratii* could delay or inhibit the dinoflagellate blooms as infected dinoflagellates become reproductively incompetent.

Key words: *Amoebophrya ceratii*, *Ceratium furca*, Mexican Pacific, parasitic dinoflagellates, red tides.

### RESUMEN

El parasitismo dentro del grupo de los dinoflagelados es un fenómeno bien documentado y presenta una amplia distribución. Los dinoflagelados del género *Amoebophrya* comúnmente parasitan a especies de dinoflagelados de vida libre, tóxicos o no tóxicos, que forman mareas rojas nocivas. Las infecciones de *Ceratium furca* causadas por *A. ceratii* se observaron en muestras de mareas rojas colectadas en la costa de Baja California, entre los 30°01'05" N, 115°51'16" W y los 31°09'33" N, 116°31'09" W. Este es el primer registro de parasitismo en

dinoflagelados en aguas del Pacífico Mexicano. Las especies causantes de esta discoloración fueron el dinoflagelado desnudo similar a *Gymnodinium*, *Ceratium furca* y *Akashiwo sanguinea*, los cuales alcanzaron concentraciones tan altas como 560 000, 762 600, y 395 400 células L<sup>-1</sup>, respectivamente. Durante el florecimiento, la temperatura del agua varió entre 13 y 17°C y su salinidad varió de 33.2 a 33.8 psu. Alrededor de 1.5% de los individuos de *C. furca* estaban infectados por el parásito intracelular *Amoebophrya ceratii*. Este parásito fue observado principalmente dentro de los especímenes de *Ceratium furca*. También se observó en *Ceratium macroceros* en muestras de red. En general, los individuos de *C. furca* manifestaron una deformidad parcial o total. Las infecciones debidas al dinoflagelado *A. ceratii* pueden llegar a retardar o inhibir los florecimientos de dinoflagelados debido a que éstos se vuelven reproductivamente incompetentes.

Palabras clave: *Amoebophrya ceratii*, *Ceratium furca*, dinoflagelados parásitos, mareas rojas, Pacífico mexicano.

## INTRODUCTION

The fact that dinoflagellates are parasites of many other protists, metazoans, and even algae was first recognized by Chatton (1912, 1952). Over 80 species of parasitic dinoflagellates have been recorded (Elbrächter & Schnepf, 1998). The recognition of such parasites requires a certain level of experience that has been lacking until now in many research groups. Dinoflagellates of the genus *Amoebophrya* are frequently parasites of marine protists including radiolaria, ciliates and other dinoflagellates (Coats et al., 1996). *Amoebophrya ceratii* (Koeppen) Cachon is a parasitic dinoflagellate broadly distributed in the northern hemisphere with infections reported from 30 host species representing 28 dinoflagellate genera from coastal waters of the Mediterranean Sea and the Atlantic and Pacific oceans (Cachon, 1964; Taylor, 1968; Elbrächter, 1973; Nishitani et al., 1985; Fritz & Nass, 1992; Coats & Bockstahler, 1994; Coats et al., 1996).

Dinoflagellate studies carried out in the North Mexican Pacific have focused mainly on the floristic and distribution of free-living toxic and non-toxic species (Hernández-Becerril, 1988; 1989; Martínez-López & Verdugo-Díaz, 2000; Gárate-Lizárraga et al., 2001). *Amoebophrya* species appear well adapted to exploit dinoflagellate blooms (Yih & Coats, 2000). Although said dinoflagellate blooms are common events along the coasts of the Baja California peninsula (Blasco, 1977; Gárate-Lizárraga et al., 2001), parasitic dinoflagellates had not been recorded. The purpose of this work is to present the first reported instance of *Amoebophrya ceratii* in these waters.

## MATERIAL AND METHODS

From June 30 to July 1, 1996, several patches of red tides were observed off the west coast of Baja California, along the northern portion of the Baja California

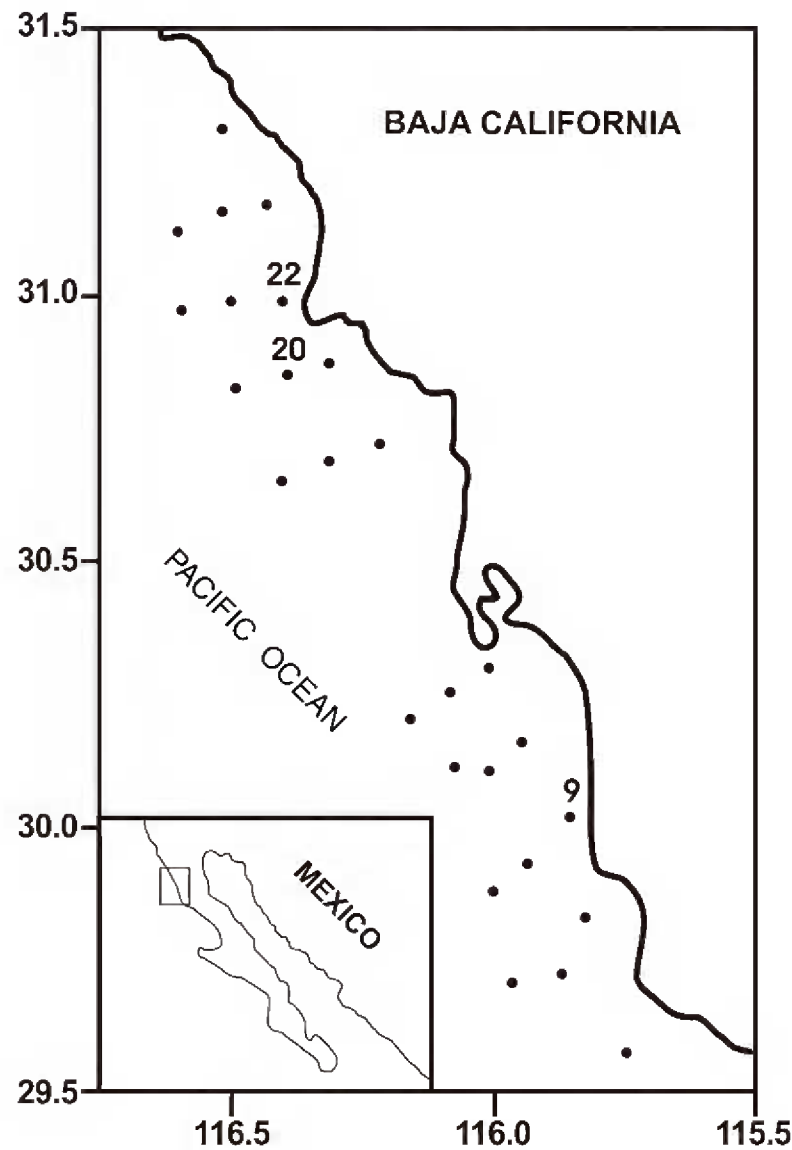


Figure 1. Location of the study area and red tides sampling stations.

peninsula, between 30°01'05" N, 115°51'16" W and 31°09'33" N, 116°31'09" W (Fig. 1). Surface samples of red tides used for this study were collected at stations 9 (samples A, B, C), 20 (A, B), and 22 (A) (Fig. 1). To support the taxonomic determinations of species surface net hauls were made at each station using a 54  $\mu$ m mesh phytoplankton net. Net samples were analysed to find other infected dinoflagellate hosts. Samples for identification and cell counts were fixed and preserved with 3% formalin, and observed in 5-ml settling chambers using a phase contrast inverted microscope (Hasle, 1978). The total phytoplankton abundance was estimated simultaneously with the species composition determinations. Nannophytoplankton (organism < 20 $\mu$ ) abundance was estimated too and nannoplankters were grouped as nannoflagellates and not identified taxonomically because of its size. Surface water temperatures were recorded using a Kalhlsico thermometer. Salinity was recorded using a CTD Inter-Ocean SO4. Microphotographs were taken with an Olympus CH-30 phase contrast microscope.

## RESULTS AND DISCUSSION

A total of 23 species of microphytoplankton were identified. Dinoflagellates were by far the most important group in abundance and in number of taxa determined (15), followed by diatoms (7) and 1 euglenophyte. Nannoflagellates and unidentified naked dinoflagellates were also important contributors to the total abundance (Table 1). Total phytoplankton abundance in the samples ranged from 128 600 to 1 636 800 cells L<sup>-1</sup>.

Table 1. Species composition and abundance (cells L<sup>-1</sup>) of phytoplankton community in the different red tide samples.

Species composition	Station 20 A	Station 22 A	Station 22 B	Station 9 A	Station 9 B	Station 9 C
<b>DIATOMS</b>						
<i>Asteromphalus heptactis</i>			200		800	800
<i>Cylindrotheca closterium</i>	400	800	2400	8400	2000	6200
<i>Chaetoceros curvisetus</i>	200		2400		8400	9400
<i>Chaetoceros lorenzianus</i>					2800	
<i>Proboscia alata</i>		800			400	400
<i>Rhizosolenia imbricata</i>	200	200			600	1200
<i>Thalassiosira</i> sp.		800				
<b>DINOFLAGELLATES</b>						
<i>Akashiwo sanguinea</i>	25000	12400	97400	157600	140000	395400
<i>Alexandrium monilatum</i>	200				3400	
<i>Ceratium furca</i>	4000	410800	762200	392800	307200	482400
<i>Ceratium fusus</i>	200	2000	1200		2400	15600
<i>Ceratium kofoidii</i>		400	400		2400	400
<i>Ceratium pentagonum</i>	400					
<i>Gonyaulax verior</i>					1200	
<i>Gymnodinium catenatum</i>				4200	1200	
<i>Gymnodinium</i> -like dinoflagellate	22400	171200	60000	341600	371200	560000
<i>Heteraulacus polyedricus</i>			800			800
Naked dinoflagellates	42800	5600	5000	8400	11600	18800
<i>Noctiluca scintillans</i>	600		800		1200	400
<i>Oxyphyxis oxytoxoides</i>			2400		800	1800
<i>Prorocentrum triestinum</i>	1200	4800	1400	4000	6400	14000
<i>Protoperidinium bipes</i>					400	



Table 1. Continuation.

Species composition	Station 20 A	Station 22 A	Station 22 B	Station 9 A	Station 9 B	Station 9 C
<i>Scrippsiella trochoidea</i>	28600	2400	400	2000	200	4200
EUGLENOPHYTES <i>Eutreptia</i> sp.	200	800				
NANNOPLANKTON Nannoflagellates	2200	56000	14600	58000	75000	125000
TOTAL ABUNDANCE	128600	669000	951600	977000	939600	1636800

Three dinoflagellate species caused the observed seawater discoloration: a *Gymnodinium*-like dinoflagellate, *Ceratium furca* (Ehrenberg) Claparède & Lachmann and *Akashiwo sanguinea* (Hirasaka) G. Hansen et Møestrup which reached concentrations as high as 560 000, 762 600, and 395 400 cells L<sup>-1</sup>, respectively. Species composition and the abundance (cells L<sup>-1</sup>) of phytoplankton community observed in the different patches are shown in Table 1. Other species that form red tides, such as *Prorocentrum triestinum* Schiller, *Scrippsiella trochoidea* (Stein) Loeblich, *Alexandrium monilatum* (Howell) F. J. R. Taylor and *Gymnodinium catenatum* Graham were also identified in the samples. *G. catenatum* had not been recorded previously off the west coast of the Baja California peninsula. During the bloom surface water temperature ranged between 13 and 17°C, while seawater salinity ranged between 33.2 and 33.8 psu.

Blooms of *A. sanguinea* have been scarcely reported in the west coast of Baja California peninsula (Turrubiates-Morales, 1994; Gárate-Lizárraga et al., 2001). However, in spite of *C. furca* being a very common species (Estrada & Blasco, 1979; Hernández-Becerril, 1989; Martínez-López & Verdugo-Díaz, 2000), no blooms of that species have been observed previously in this area. Isolated blooms of *C. furca* have been reported in coastal lagoons (Gárate-Lizárraga & Siqueiros-Beltrones, 1998). Highest values of biomass (chlorophyll a) during the blooms observed in this study varied from 9.6 to 16.5 mg/m<sup>3</sup> (Gárate-Lizárraga & Martínez-López, 1996). These blooms are responsible for local increases in the phytoplankton biomass, and seem to be very important in terms of the fertility of this coastal zone.

About 1.5% of the individuals of *C. furca* observed were infected by the intracellular parasitic dinoflagellate *Amoebophyra ceratii* (Koeppen) Cachon. This

parasite was observed mainly inside specimens of *Ceratium furca*. In net samples, few specimens of *Ceratium macroceros* (Ehrenberg) Vanhöffen were infected too. In general, infected individuals of *C. furca* were partially or totally deformed (Fig. 2). Elbrächter (1973) reported the first infections of *C. furca* by *A. ceratii* for German waters. Measurements made on 15 infected specimens (114 - 136  $\mu\text{m}$ ) and 15 healthy ones (96 - 120  $\mu\text{m}$ ) showed that the former had a mean length/wide ratio of 2.67 vs. a 3.3 ratio for the latter. The other red tide causing dinoflagellates recorded were not infected by *A. ceratii*.

All *Amoebophrya* are intracellular parasites and infections occur when dinospores, the biflagellated dispersal stage of the parasite, attach to the outer membrane and penetrate into the cytoplasm of the host. Inside the host, the parasite differentiates into trophont (vegetative) stage which soon increases in size and begins a series of nuclear divisions. It differentiates into an episome which sinks progressively in the hyposome (Cachon, 1964; Coats & Bockstahler, 1994). Nuclear divisions are not followed by cytoplasmic divisions. Thus, trophonts of late infections are large polynucleated organisms later becoming multiflagellate that occupy most of host cell.

The ecological importance of parasitic dinoflagellates is particularly evident during epidemic outbreaks that cause mass mortality of host organisms. Yih & Coats (2000) have pointed out that *Amoebophrya* species appear well adapted to exploit dinoflagellate blooms in enriched coastal environments because they may infect up to 80% of the potential hosts (Coats, 1999). The bloom of *C. furca* here reported

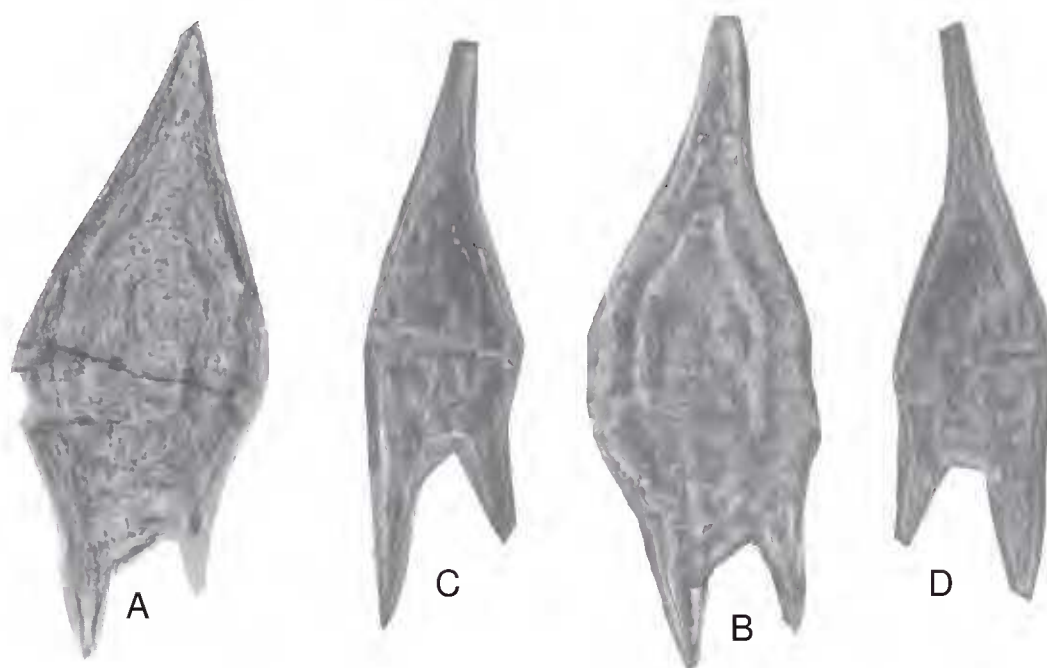


Figure 2. Light microscopy photograph of *Ceratium furca*: A and B = *C. furca* infected by *Amoebophrya ceratii*; A = typical view of *A. ceratii* resembling a "fingerprint"; C and D = normal specimens.

occurred in an upwelling zone (Torres-Moye et al., 1986). Occurrence of dinoflagellate blooms are common events in the Baja California peninsula coasts and have been linked to the nutrient enriched waters of upwelling events (Blasco, 1977; Estrada & Blasco, 1979; Packard et al., 1978; Gárate-Lizárraga et al., 2001). Thus, the possibility of infections by *A. ceratii* are high. Infections by *A. ceratii* delay or inhibit dinoflagellate blooms because infected dinoflagellates become reproductively incompetent (Elbrächter, 1973; Nishitani et al., 1985).

Most events of *A. ceratii* infections have been observed during red tides (Taylor 1968; Elbrächter, 1973; Fritz & Nass, 1992; Coats & Bockstahler, 1994; Coats et al., 1996). Taylor (1968) has suggested that *A. ceratii* could be used as an effective biological control against the development of dense dinoflagellate blooms. Albeit, Nishitani et al. (1985) argued that *Amoebophrya* would not be a good biological control because it was not host specific. In vitro observations by Coats et al. (1996) and Coats & Park (2002) showed that cultures of *A. ceratii* failed to infect several other dinoflagellate species, e.g. *A. sanguinea* and *Gymnodinium instriatum* (Fraudenthal et Lee) Coats, and suggested that this taxon represents a species complex composed of several host-specific parasites, and that Taylor's suggestion should be given more consideration.

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